

Monitoring Process Effectiveness

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Treatment of municipal sludges to produce biosolids which meet federal and/or state requirements for land application requires process monitoring. The goal of process monitoring is to produce biosolids of consistent and reliable quality. In its simplest form, for Class B treatments, this may be monitoring the pH of an alkaline treatment or determining flow rates, temperatures and volatile solids reduction of mesophilic anaerobic or aerobic digesters. However, to achieve Class A, process monitoring includes observation and recording process conditions as well as monitoring indicator microorganisms or specific pathogens (U.S. EPA, 2003).

Operation of conventional municipal sludge treatment systems is well documented (WEF MOP, 1998; U.S. EPA, 1979, U.S. EPA, 2003). Process monitoring parameters are relatively well defined for these systems. However, the design and operation of many systems are customized to meet facility needs and requirements, and may include innovative processes which are relatively new and in some cases are available only through a single technology vendor. In these situations provisions must be made for defining those parameters which affect process effectiveness and to design a monitoring plan or system which will ensure that biosolids are produced of a known and acceptable quality. Such process monitoring is an essential and important part of producing a biosolids of acceptable quality; however the focus of this manuscript is on monitoring the effectiveness of biosolids treatment processes based upon assaying the product.

According to the *NIST/SEMATECH e-Handbook of Statistical Methods*, (NIST/SEMATECH, 2006), the purpose of statistical quality control is to ensure, in a cost efficient manner, that the product shipped to customers (in this case biosolids) meets their specifications (regulatory requirements, nutrients, aesthetics). Inspecting every product is costly and inefficient, but the consequences of shipping non conforming product can be significant in terms of customer dissatisfaction. Statistical Quality Control is the process of inspecting enough product from given lots to probabilistically ensure a specified quality level. Therefore, in order to ensure that a treatment process is producing biosolids of known quality, routine monitoring of the biosolids is reasonable and required. Table 1, displays the minimum sampling frequency which is required for land applied biosolids (40 CFR 503.16). Note that this sampling schedule is intended for demonstrating regulatory compliance based upon the mass of biosolids produced at a given facility. It is not intended to meet the rigor of a well designed quality control program.

Table 1. Minimum sampling frequency for land application of biosolids.

Amount of sewage sludge¹(metric tons per 365 day period)	Frequency
Greater than zero but less than 290	Once per year.
Equal to or greater than 290 but less than 1,500	Once per quarter (four times per year).
Equal to or greater than 1,500 but less than 15,000	Once per 60 days (six times per year).
Equal to or greater than 15,000	Once per month (12 times per year).

¹Either the amount of bulk sewage sludge applied to the land or the amount of sewage sludge prepared for sale or give-away in a bag or other container for application to the land (dry weight basis).

Fecal coliform densities have historically been used to demonstrate the disinfection efficiency of biosolids treatment processes (Yanko, 1981; Meckes, et al, 1998). Indeed, current federal regulations (40 CFR 503 Subpart D) specifically require facilities producing Class A biosolids to determine (at a minimum) fecal coliform, or *Salmonella* sp. densities prior to land application. However, there are concerns that fecal coliforms may not be the best indicator for evaluating process effectiveness. For example, recently Higgins, et al. (2008) demonstrated that thermophilic anaerobic digestion followed by centrifuge dewatering could result in high densities of *Escherichia coli* (*E. coli*). This species is the predominant member of the fecal coliform group of organisms found in municipal sludges and biosolids. However, in that same study, selected species of pathogenic bacteria were not observed in the biosolids product. This suggests that, at least with respect to thermophilic digestion followed by centrifuge dewatering, *E. coli*, (therefore fecal coliforms) is a poor indicator of process/disinfection effectiveness. Additionally, coliform organisms are not necessarily good indicators of a processes ability to reduce the number of viruses, protozoans, or helminthes from municipal sludge. Consequently, federal regulations require that facilities must analyze biosolids for enteric viruses and viable helminth ova when using processes which have not been treated by defined systems (Class A, Alternative 3 or 4, 40 CFR 503). Although quantification of enteric viruses and viable helminth ova in treated biosolids provides assurance that densities of these organisms may be less than detection limits, such results may provide little or no information regarding process effectiveness when densities of these organisms are low (or below detection limits) prior to treatment. Finding low densities of viable helminth ova in sludges throughout the northern U.S. is not unusual (O'Donnell, et al., 1984). A similar situation has also been noted for recoverable enteric viruses in wastewaters (Melnick, et al., 1995). In such cases, it is not reasonable to gauge the effectiveness of a process by simply determining the density of a microorganism in treated biosolids.

As noted above, under specific conditions, Federal regulations require facilities to evaluate sludges and biosolids for bacteria, viruses and viable helminth ova. One of these conditions is when one demonstrates PFRP (Process to Further Reduce Pathogens) equivalency. In order to demonstrate that a sludge treatment process is capable of reducing/eliminating enteric viruses and viable helminth ova, the densities of these organisms are determined before and after treatment. PFRP equivalent processes must demonstrate a minimum of 99.9% reduction in the number of enteric viruses (as measured by cell culture) while achieving a minimum reduction of 99% of the viable helminth ova. Consequently, prior to such demonstrations sludges with relatively low densities of enteric viruses and viable helminth ova must be augmented (or spiked) with these organisms to ensure that organism densities are sufficient to demonstrate process effectiveness. For a more complete discussion of the equivalency determination process please see: <http://www.epa.gov/ORD/NRMRL/pec>. Process operating conditions must be closely monitored when demonstrating PFRP equivalency. The conditions under which disinfection is achieved are then used to define minimum process operating conditions. For example, the PFRP definition for windrow composting states that the process must be operated to maintain a minimum temperature of 55° C for a minimum of 15 days. Furthermore, during the time that the temperature of the compost is $\geq 55^{\circ}$ C the windrow must be turned a minimum of five times. Process monitoring must then include temperature measurements over the processing period for each batch treated. Yanko (1987) showed that when operated under these conditions enteric viruses and viable helminth ova were not detected. He also noted that fecal coliform densities were reduced to below 1000/g (dry weight) and *Salmonella* sp. were rarely detected. As a consequence of such work, as long as specified operating conditions are achieved and appropriately monitored, a given process should consistently reduce pathogens to below detection limits. Monitoring the density of indicator organisms such as fecal coliforms following treatment is used to demonstrate that such processes can consistently reduce the density of enteric microorganisms.

Fecal coliforms are well known as indicators of polluted water (APHA, 2005). One of the biggest advantages of using these organisms as microbial indicators is that they are consistently found in high densities in municipal sludges. Since these organisms are most often benign, are relatively easy to enumerate, and respond to treatment similar to known pathogenic strains (Meckes, et al. 1998; Meckes and Rhodes, 2004) it is reasonable to use them for determining process effectiveness. However, other organisms or groups of organisms share many or all of the traits which make fecal coliform a good choice for monitoring process effectiveness. For example, enterococci are a group of Gram positive bacteria which inhabit the intestines of healthy individuals (APHA, 2005). Consequently they are commonly found in relatively high densities in municipal wastewaters and sludges. Enterococci are most often benign, relatively easy to enumerate and they appear to respond to treatment similar to known pathogenic strains. However, they are not routinely enumerated because they are not used for compliance monitoring. A larger group of organisms are the heterotrophic bacteria. This group includes fecal coliforms as well as any other bacteria which can utilize simple carbohydrates as a source of nutrition and will form a colony on a defined semi-solid media within a specified time frame when incubated at a specified temperature. The

number of heterotrophic bacteria in sludge is two to three orders of magnitude greater than the number of fecal coliforms. This suggests that this broad and diverse group of bacteria may be a better overall indicator of process effectiveness since it is likely that this group will include bacteria which are more resistant to sludge treatments than potentially pathogenic strains (the converse would also apply).

Bacteria which produce endospores are able to survive environmental stressors better than other types of bacteria (Rice, et al., 1996). These include anaerobic bacteria such as *Clostridia* sp. and aerobic bacteria such as *Bacillus* sp. Endospores of these organisms would be poor indicators of process effectiveness for most biological treatments since there may be insufficient stress placed on these organisms to induce the formation of endospores. However, they may be useful to monitor process effectiveness for physical/chemical treatment systems, and in some cases, may be a good indicator of overall treatment (Meckes and Rhodes, 2004).

Protozoan parasites such as *Cryptosporidium parvum* and *Giardia lamblia* have been identified in municipal wastewaters (Chauret, et al., 1999). However, the ability of these protozoans to survive conventional and innovative municipal sludge treatment processes has not been widely studied. This is most likely due to problems associated with analysis. Helminth ova have routinely been used as indicators of treatment process effectiveness (Yanko, 1987, U.S. EPA, 2003). Ova of the helminth, *Ascaris suum* is relatively robust and sufficiently large so that it can be isolated from sludges and will survive environmental stressors (O'Donnell, et al., 1984).

The analytical techniques used to enumerate enteric viruses in municipal sludges are tedious, time consuming and expensive. This is largely due to the need for maintaining a cell (tissue) culture which is used as host for the virus (APHA, 2005; U.S. EPA, 2003). Coliphage are a group of bacterial viruses which infect *E. coli* which have been used for process monitoring (Moce'-Llivina, et al., 2003; Nappier, et al., 2006). These viruses are similar to animal viruses however, because they infect bacteria, assay costs are much lower.

It should be clear from the above discussion that there are many indicator organisms and frank pathogens which could be used for monitoring process effectiveness. Monitoring fecal coliform densities is attractive because of the established compliance monitoring requirement. However, the limitations of using fecal coliforms for process monitoring have been noted. Furthermore there is no necessity for using the same indicator for compliance and process monitoring. When selecting a process indicator one should base the selection on several factors. These include: representativeness; response time; and cost. Depending on the treatment process, some indicator organisms appear to be more reliable than others. For example, processes which use alkaline treatments above pH 12 along with heat (temperatures in excess of 50°C) are effective in reducing the numbers of bacterial endospores (Meckes, and Rhodes, 2004), however there is no evidence which would support the uses of such indicator organisms for biological or other types of thermal treatment systems. Response time is the time between collection of process samples and assay results. For batch treatments this would be the amount of

holding time needed for accepting or rejecting the batch, therefore it relates directly to the amount of space available for storage of product. Viability tests for helminth ova take a minimum of two weeks, and tissue culture assays for enteric viruses may require a similar amount of time for verification. Consequently, response times when using these organisms for process monitoring are excessive. Many of the bacteria and bacteriophage assays discussed above may be completed within one day, making them more attractive as indicators of process effectiveness. However, it is important to note that when considering the use of bacteria or bacteriophage as process indicators, the process being evaluated must have a demonstrated ability to reduce enteric viruses and viable helminth ova numbers to below regulatory limits. Finally, the cost associated with monitoring process indicators should be low. Low analytical costs are desirable not only to limit the cost of processing, but to promote frequent product testing. Here again, assays for bacteria are less expensive than those required for enteric virus or viable helminth ova.

Summary:

Monitoring process effectiveness is not only required, it is good practice. Process monitoring must include observation and recording of process operating conditions as well as measuring product quality. Microbiological indicators of process effectiveness include bacteria, viruses and parasites, however selection of appropriate process effectiveness indicators favors the use of bacterial indicators.

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